Supplementary Material

A comparative study on the quality of protein crystals obtained using the cross-diffusion microbatch and sitting-drop vapor diffusion methods

Hai Hou ^{a, ‡}, Bo Wang ^{a, ‡}, Shan-Yang Hu ^a, Jing-Zhang Wang ^b, Peng-Fei Zhu ^a, Yue Liu ^a, Meng-Ying Wang ^a, and Da-Chuan Yin ^{a, *}

^aKey Laboratory for Space Bioscience & Biotechnology, School of Life Sciences, Northwestern Polytechnical University, Xi'an 710072, Shaanxi, P. R. China ^bDepartment of Medical Technology, Affiliated Hospital, College of Medicine, Hebei Univers ity of Engineering, Handan 056002, Hebei, P. R. China

Supplementary Figure S1. Comparison of images of the crystallization plate using the CDM method and the SDVD method. (a) Image of crystallization plate using the traditional sitting

drop vapor-diffusion (SDVD) method. (b) Image of crystallization plate using the crossdiffusion microbatch (CDM) method. The size of this plate is made to be compatible with the SBS standard plate. This crystallization plate is similar to the traditional SDVD plate, except for the material and the reservoir, which was replaced by more pits to increase the number of protein concentrations or pH gradients.



Supplementary Figure S2. (a) A comparison of crystallization screening hits for the 11 proteins using CDM and the traditional sitting drop vapor-diffusion method. (b) A statistical

Abbreviations: CDM, cross-diffusion microbatch; SDVD, sitting-drop vapor-diffusion.

analysis of the average screening hits between the CDM method and the traditional method (the error bars show the standard error of the mean; n = 11). The number of hits was normalized based on the data for the CDM method. The results demonstrated an extremely significant difference between the two groups (n = 11, P = 0.00026, *i.e.* <0.01). The CDM method clearly increased the number of crystallization hits compared with the traditional sitting drop vapor-diffusion method.



Supplementary Figure S3. Crystal morphology comparison between the CDM and SDVD crystallization plates. CDM: the crystals were optically better in the CDM plates than in the

SDVD plates. SDVD: the crystals were optically better in the SDVD plates than in the CDM plates. Comparable: the optical perfection of the crystals was comparable in the two plates. The results showed that more crystals grown in the CDM plates exhibited better optical perfection than those grown in the SDVD plates. The following criteria were used to compare the optical perfection: First, the crystals were categorized into different groups and ranked as follows: single crystal > single crystal with cracks > twin crystal > needle cluster > quasi-crystal. If the crystals were in the same group, then they were compared according to their size: larger crystals were defined as more optically perfect. If the crystal sizes were similar (size difference was within 5%), they were optically comparable.



Supplementary Figure S4. (a) A statistical comparison of the mosaicity of the top 5 proteinase K crystals after normalization. However, for mosaicity, the results demonstrated

no significant difference between the two groups (n = 5, P = 0.537, *i.e.*, >0.05). (b) A statistical comparison of the mosaicity of the top 5 lysozyme crystals after normalization. Nevertheless, for the mosaicity, the difference between the two groups was not significant (n = 5, P = 0.274, *i.e.*, >0.05).



	Diffraction data statistics of proteinase K crystals in CDM crystallization plate								
?									
	Crystal 1	Crystal 2	Crystal 3	Crystal 4	Crystal 5				
	50-1.54	50-1.54	50-1.65	50-1.66	50-1.66				
Resolution range (Å)	(1.57-1.54)	(1.57-1.54)	(1.68-1.65)	(1.69-1.66)	(1.69-1.66)				
Mosaicity (°)	0.47	0.42	0.60	0.61	0.46				
< <i>I>/<</i> σ(<i>I</i>)>	25.91 (7.89)	20.65 (3.39)	15.58 (2.15)	16.27 (2.76)	13.60 (2.21)				
Wilson Plot B factor	6.50	8.00	7.40	9.70	7.60				
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2				
	<i>a</i> = 68.17	a = 67.74	a = 66.52	a = 68.05	a = 67.75				
Cell dimensions	<i>b</i> = 68.17	b = 67.74	h = 66.52	b = 68.05	b = 67.75				
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 102.13	c = 101.81	c = 99.51	c = 102.34	c = 100.56				
$\alpha,\beta,\gamma(^\circ)$	$\alpha = \beta = \gamma =$	$\alpha = \beta = \gamma = 90$							
	90	ар <i>ү</i> У б	ω <i>μ</i> γ 50	арууб 1	ар <i>у</i> у у о				
Exposure time (min)			5						
Oscillation angle (°)			1						
Detector distance			150						
(mm)			150						
Wavelength (Å)			1.54179						

Supplementary Table S1. X-ray diffraction data statistics of 23 proteinase K crystals in CDM crystallization plate in same size

Values in parentheses are for the highest resolution shell.

Diffraction data statistics of proteinase K crystals in CDM crystallization plate ? Crystal 6 Crystal 7 Crystal 8 Crystal 9 Crystal 10 50-1.66 50-1.66 50-1.66 50-1.73 50-1.76 Resolution range (Å) (1.69-1.66) (1.69 - 1.66)(1.69 - 1.66)(1.76 - 1.73)(1.79 - 1.76)Mosaicity (°) 0.41 0.45 0.64 0.53 0.46 14.53 (2.44) 19.49 (4.50) 19.12 (2.70) 12.06 (2.09) <*I*>/<*σ*(*I*)> 11.79 (2.04) Wilson Plot B 9.90 8.40 9.80 11.10 8.30 factor $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ P4₃2₁2 Space group $P4_{3}2_{1}2$ a = 67.72a = 67.74*a* = 68.02 *a* = 67.63 a = 67.80Cell dimensions b = 67.72b = 67.74*b* = 68.02 b = 67.63b = 67.80*a*, *b*, *c* (Å) c = 101.90c = 102.26c = 101.83c = 101.72c = 102.34 α, β, γ (°) $\alpha = \beta = \gamma = 90$ Exposure time 5 (min) Oscillation angle 1 (°) Detector distance 150 (mm) Wavelength (Å) 1.54179

Continued Supplementary Table S1

Diffraction data statistics of proteinase K crystals in CDM crystallization plate ? Crystal 11 Crystal 12 Crystal 13 Crystal 14 Crystal 15 50-1.78 50-1.82 50-1.83 50-1.84 50-1.85 Resolution range (Å) (1.81-1.78) (1.85 - 1.82)(1.86 - 1.83)(1.87 - 1.84)(1.88 - 1.85)0.57 0.50 0.93 Mosaicity (°) 0.58 0.38 14.93 (2.09) <*I*>/<*σ*(*I*)> 13.29 (2.26) 11.54 (2.23) 9.70 (2.31) 13.96 (2.01) Wilson Plot B 13.00 12.90 11.10 8.30 11.00 factor $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ P4₃2₁2 Space group $P4_{3}2_{1}2$ a = 67.54a = 67.41a = 67.80a = 67.72*a* = 67.88 Cell dimensions b = 67.54b = 67.41b = 67.80b = 67.72*b* = 67.88 *a*, *b*, *c* (Å) c = 101.72c = 100.98c = 102.44c = 102.22c = 101.49 α, β, γ (°) $\alpha = \beta = \gamma = 90$ Exposure time 5 (min) Oscillation angle 1 (°) Detector distance 150 (mm) Wavelength (Å) 1.54179

Continued Supplementary Table S1

Diffraction data statistics of proteinase K crystals in CDM crystallization plate ? Crystal 16 Crystal 17 Crystal 18 Crystal 19 Crystal 20 50-1.86 50-1.87 50-1.88 50-1.93 50-1.99 Resolution range (Å) (2.02-1.99) (1.89 - 1.86)(1.90 - 1.87)(1.91 - 1.88)(1.96 - 1.93)1.00 0.51 Mosaicity (°) 0.42 0.63 0.44 20.22 (4.16) 14.51 (2.07) <*I*>/<*σ*(*I*)> 10.72 (2.16) 7.95 (2.15) 7.91(2.01) Wilson Plot B 12.00 10.30 13.00 12.80 18.60 factor $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ P4₃2₁2 Space group $P4_{3}2_{1}2$ a = 67.99a = 67.76a = 67.78a = 67.73a = 67.77Cell dimensions b = 67.99b = 67.76b = 67.78b = 67.73b = 67.77*a*, *b*, *c* (Å) c = 101.83c = 101.53c = 102.82c = 101.65c = 102.51 α, β, γ (°) $\alpha = \beta = \gamma = 90$ Exposure time 5 (min) Oscillation angle 1 (°) Detector distance 150 (mm) Wavelength (Å) 1.54179

Continued Supplementary Table S1

	Diffraction data statistics of proteinase K crystals in						
?	(CDM crystallization	plate				
	Crystal 21	Crystal 22	Crystal 23				
Resolution range (Å)	50-2.17	50-2.18	50-2.20				
	(2.21-2.17)	(2.22-2.18)	(2.24-2.20)				
Mosaicity (°)	0.98	0.80	0.49				
<1>/<\sigma(1)>	8.40 (2.65)	7.26 (2.30)	9.06 (2.22)				
Wilson Plot B factor	13.00	12.30	17.40				
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2				
	<i>a</i> = 67.84	<i>a</i> = 67.87	<i>a</i> = 67.59				
Cell dimensions	<i>b</i> = 67.84	<i>b</i> = 67.87	<i>b</i> = 67.59				
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 101.79	<i>c</i> = 102.21	<i>c</i> = 101.46				
α, β, γ (°)	$\alpha = \beta = \gamma =$ 90	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$				
Exposure time (min)		5					
Oscillation angle (°)		1					
Detector distance (mm)		150					
Wavelength (Å)		1.54179					

Supplementary	Table	S2.	X-ray	diffraction	data	statistics	of	20	proteinase	Κ	crystals	in
SDVD crystalli	zation j	plate	in sam	e size								

	Diffracti	ion data statistics of proteinase K crystals in SDVD crystallization plate						
2	Crystal 1	Crystal 2	Crystal 3	Crystal 4	Crystal 5			
	50-1.66	50-1.66	50-1.70	50-1.72	50-1.73			
Resolution range (A)	(1.69-1.66)	(1.69-1.66)	(1.70-1.73)	(1.75-1.72)	(1.76-1.73)			
Mosaicity (°)	0.66	0.46	0.51	0.57	0.53			
< <i>I>/<</i> σ(<i>I</i>)>	20.96 (3.32)	17.63 (2.70)	13.45 (2.04)	13.65 (2.13)	11.92 (2.19)			
Wilson Plot B factor	13.30	11.50	11.90	10.40	11.00			
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2			
	<i>a</i> = 68.10	<i>a</i> = 68.12	<i>a</i> = 67.93	<i>a</i> = 67.82	<i>a</i> = 67.43			
Cell dimensions	<i>b</i> = 68.10	<i>b</i> = 68.12	<i>b</i> = 67.93	<i>b</i> = 67.82	<i>b</i> = 67.43			
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 101.90	<i>c</i> = 102.33	<i>c</i> = 101.62	<i>c</i> = 101.41	<i>c</i> = 101.00			
α, β, γ (°)	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$			
Exposure time (min)			5					
Oscillation angle (°)			1					
Detector distance (mm)			150					
Wavelength (Å)			1.54179					

Diffraction data statistics of proteinase K crystals in SDVD crystallization plate ? Crystal 6 Crystal 7 Crystal 8 Crystal 9 Crystal 10 50-1.77 50-1.78 50-1.85 50-1.86 50-1.87 Resolution range (Å) (1.80 - 1.77)(1.81 - 1.78)(1.88 - 1.85)(1.89 - 1.86)(1.90 - 1.87)0.61 Mosaicity (°) 0.79 0.75 1.28 0.78 14.45 (2.07) 13.07 (2.08) 10.34 (3.63) 7.85 (2.07) <*I*>/<*σ*(*I*)> 17.62 (2.58) Wilson Plot B 12.40 10.60 14.40 19.60 11.30 factor $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ P4₃2₁2 Space group $P4_{3}2_{1}2$ *a* = 68.19 a = 67.72a = 67.90*a* = 67.32 *a* = 67.98 Cell dimensions b = 68.19b = 67.72b = 67.90b = 67.32*b* = 67.98 *a*, *b*, *c* (Å) c = 100.50c = 102.04c = 95.15c = 102.17c = 101.88 α, β, γ (°) $\alpha = \beta = \gamma = 90$ Exposure time 5 (min) Oscillation angle 1 (°) Detector distance 150 (mm) Wavelength (Å) 1.54179

Continued Supplementary Table S2

	Diffraction data statistics of proteinase K crystals in SDVD crystallization plate								
2	Cructal 11	Crystal 12	Crystal 12	Crystal 14	Crystal 15				
	Crystal 11	Crystal 12	Crystar 15	Crystal 14	Crystal 15				
Resolution range	50-1.95	50-1.98	50-2.05	50-2.08	50-2.09				
(Å)	(1.98-1.95)	(2.01-1.98)	(2.09-2.05)	(2.12-2.08)	(2.13-2.09)				
Mosaicity (°)	0.69	1.39	0.83	1.12	1.61				
<i>/<\sigma(I)></i>	9.58 (2.07)	11.82 (2.33)	6.10 (2.04)	10.85 (2.17)	11.18 (2.13)				
Wilson Plot B factor	11.60	18.30	15.90	16.40	10.10				
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2				
Cell dimensions	<i>a</i> = 67.75	<i>a</i> = 67.75	<i>a</i> = 67.95	<i>a</i> = 66.66	<i>a</i> = 67.59				
	<i>b</i> = 67.75	<i>b</i> = 67.75	<i>b</i> = 67.95	<i>b</i> = 66.66	<i>b</i> = 67.59				
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 101.21	<i>c</i> = 101.58	<i>c</i> = 101.61	<i>c</i> = 100.10	<i>c</i> = 100.84				
α, β, γ (°)	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$				
Exposure time			5						
(min)									
Oscillation angle			1						
(°)									
Detector distance			150						
(mm)									
Wavelength (Å)			1.54179						

Diffraction data statistics of proteinase K crystals in SDVD crystallization plate ? Crystal 16 Crystal 17 Crystal 18 Crystal 19 Crystal 20 50-2.11 50-2.20 50-2.38 50-2.78 50-3.07 Resolution range (Å) (2.15-2.11) (2.24 - 2.20)(2.42 - 2.38)(2.83 - 2.78)(3.12 - 3.07)0.81 0.39 0.48 Mosaicity (°) 0.47 0.38 9.62 (7.99) 5.34 (2.40) 5.09 (2.31) <*I*>/<*σ*(*I*)> 5.14 (2.09) 3.96 (2.96) Wilson Plot B 16.00 17.70 17.70 17.30 21.20 factor $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ $P4_{3}2_{1}2$ P4₃2₁2 Space group $P4_{3}2_{1}2$ *a* = 68.03 a = 67.72a = 67.84a = 67.73a = 67.69Cell dimensions *b* = 68.03 b = 67.72b = 67.84b = 67.73*b* = 67.69 *a*, *b*, *c* (Å) c = 102.80c = 102.11c = 102.63c = 102.59c = 102.37 α, β, γ (°) $\alpha = \beta = \gamma = 90$ Exposure time 5 (min) Oscillation angle 1 (°) Detector distance 150 (mm) Wavelength (Å) 1.54179

Continued Supplementary Table S2

	Diffraction data statistics of lysozyme crystals in CDM crystallization plate									
?										
	Crystal 1	Crystal 2	Crystal 3	Crystal 4	Crystal 5					
	50-1.66	50-1.66	50-1.78	50-1.83	50-1.86					
Resolution range (Å)	(1.69-1.66)	(1.69-1.66)	(1.81-1.78)	(1.86-1.83)	(1.89-1.86)					
Mosaicity (°)	0.60	0.57	0.83	0.52	0.74					
<i>/<\sigma(I)></i>	23.18 (8.58)	24.29 (2.11)	21.92 (2.24)	15.85 (2.02)	19.98 (2.12)					
Wilson Plot B factor	16.40	14.00	18.20	19.40	14.10					
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2					
	<i>a</i> = 76.91	<i>a</i> = 78.22	<i>a</i> = 78.02	<i>a</i> = 76.47	<i>a</i> = 76.71					
Cell dimensions	<i>b</i> = 76.91	<i>b</i> = 78.22	<i>b</i> = 78.02	<i>b</i> = 76.47	<i>b</i> = 76.71					
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 38.35	<i>c</i> = 37.62	<i>c</i> = 37.55	<i>c</i> = 37.58	<i>c</i> = 37.32					
α, β, γ (°)	$\alpha = \beta = \gamma =$ 90	$\alpha = \beta = \gamma = 90$								
Exposure time (min)			5							
Oscillation angle (°)			1							
Detector distance (mm)			150							
Wavelength (Å)			1.54179							

Supplementary Table S3. X-ray diffraction data statistics of 27 lysozyme crystals in CDM crystallization plate in same size

	Diffraction data statistics of lysozyme crystals in CDM crystallization plate							
2	Crystal 6	Crystal 7	Crystal 8	Crystal 9	Crystal 10			
Resolution range	50-1.86	50-1.86	50-1.86	50-1.86	50-1.86			
(Å)	(1.89-1.86)	(1.89-1.86)	(1.89-1.86)	(1.89-1.86)	(1.89-1.86)			
Mosaicity (°)	0.59	0.82	0.84	0.89	0.52			
< <i>I>/<</i> σ(<i>I</i>)>	18.70 (3.34)	18.75 (2.12)	15.50 (3.05)	22.0 (2.46)	12.93 (3.67)			
Wilson Plot B factor	18.90	15.00	14.70	16.90	20.70			
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2			
Call dimensions	<i>a</i> = 76.80	<i>a</i> = 76.47	<i>a</i> = 76.91	<i>a</i> = 76.77	<i>a</i> = 76.82			
	<i>b</i> = 76.80	<i>b</i> = 76.47	<i>b</i> = 76.91	<i>b</i> = 76.77	<i>b</i> = 76.82			
a, b, c (A)	<i>c</i> = 37.35	<i>c</i> = 37.58	<i>c</i> = 37.74	<i>c</i> = 37.27	<i>c</i> = 37.39			
α, β, γ (°)	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$			
Exposure time			5					
(min)								
Oscillation angle			1					
(°)								
Detector distance (mm)			150					
Wavelength (Å)			1.54179					

	Diffraction data statistics of lysozyme crystals in CDM crystallization plate							
2	Crystal 11	Crystal 12	Crystal 13	Crystal 14	Crystal 15			
Resolution range	50-1.87	50-1.92	50-1.93	50-1.94	50-1.94			
(Å)	(1.90-1.87)	(1.95-1.92)	(1.96-1.93)	(1.97-1.94)	(1.97-1.94)			
Mosaicity (°)	0.83	1.03	0.50	0.86	0.86			
< <i>I>/<</i> σ(<i>I</i>)>	17.73 (2.89)	16.84 (2.44)	10.85 (2.02)	18.37 (2.145)	14.36 (2.56)			
Wilson Plot B factor	22.20	20.20	21.40	25.30	24.10			
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2			
	<i>a</i> = 77.24	<i>a</i> = 76.57	<i>a</i> = 76.67	<i>a</i> = 77.27	<i>a</i> = 76.43			
Cell dimensions	<i>b</i> = 77.24	<i>b</i> = 76.57	<i>b</i> = 76.67	<i>b</i> = 77.27	<i>b</i> = 76.43			
<i>a</i> , <i>b</i> , <i>c</i> (A)	<i>c</i> = 37.81	<i>c</i> = 37.08	<i>c</i> = 37.20	<i>c</i> = 37.07	<i>c</i> = 36.74			
α, β, γ (°)	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$			
Exposure time			5					
(min)								
Oscillation angle			1					
(°)								
Detector distance (mm)			150					
Wavelength (Å)			1.54179					

	Diffraction data statistics of lysozyme crystals in CDM crystallization plate								
?									
	Crystal 16	Crystal 17	Crystal 18	Crystal 19	Crystal 20				
	50-2.02	50-2.03	50-2.08	50-2.09	50-2.10				
Resolution range (Å)									
	(2.05-2.02)	(2.06-2.03)	(2.12-2.08)	(2.13-2.09)	(2.14-2.10)				
Mosaicity (°)	1.08	0.61	0.52	1.01	0.70				
Mosuloty ()	1.00	0.01	0.02	1.01	0.70				
< <i>I>/<</i> \sigma(<i>I</i>)>	14.49 (2.30)	18.16 (3.23)	13.86 (2.14)	17.36 (2.01)	21.14 (2.30)				
		•• ••	•• ••	40.00	• • • •				
Wilson Plot B factor	14.70	23.20	23.60	19.20	21.40				
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2				
	<i>a</i> = 77.00	a = 78.30	a = 77.29	a = 76.09	a = 7879				
Cell dimensions	b = 77.00	<i>u</i> 70.50	u 11.29	<i>u</i> 70.09	<i>u</i> 10.19				
	0 - 77.00	<i>b</i> = 78.30	<i>b</i> = 77.29	<i>b</i> = 76.09	<i>b</i> = 78.79				
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 37.70								
		<i>c</i> = 35.93	<i>c</i> = 37.04	<i>c</i> = 37.17	c = 35.41				
α, ρ, γ ()	$\alpha = \beta = \gamma =$	$\alpha = \beta = \gamma = 90$							
	90	, ,	, ,	, ,	, ,				
Francisco di esca (ancia)			F						
Exposure time (min)			5						
Oscillation angle (°)			1						
Detector distance			150						
(mm)									
Wavelength (Å)			1 54170						
wavelengui (A)			1.341/7						

	Diffraction data statistics of lysozyme crystals in CDM crystallization plate								
2	Crystal 21	Crystal 22	Crystal 23	Crystal 24	Crystal 25				
Resolution range	50-2.12	50-2.12	50-2.21	50-2.23	50-2.23				
(Å)	(2.16-2.12)	(2.16-2.12)	(2.25-2.21)	(2.27-2.23)	(2.27-2.23)				
Mosaicity (°)	1.03	1.18	0.96	0.62	0.62				
< <i>I>/<</i> σ(<i>I</i>)>	15.56 (2.17)	24.25 (2.38)	22.49 (8.30)	10.32 (2.09)	11.40 (2.35)				
Wilson Plot B factor	21.40	22.60	19.80	27.10	25.40				
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2				
Call dimensions	<i>a</i> = 75.72	<i>a</i> = 76.12	<i>a</i> = 77.99	<i>a</i> = 76.66	<i>a</i> = 77.66				
	<i>b</i> = 75.72	<i>b</i> = 76.12	<i>b</i> = 77.99	<i>b</i> = 76.66	<i>b</i> = 77.66				
a, b, c (A)	<i>c</i> = 36.83	<i>c</i> = 37.18	<i>c</i> = 36.86	<i>c</i> = 37.51	<i>c</i> = 37.46				
α, β, γ (°)	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$				
Exposure time			5						
(min)									
Oscillation angle			1						
(°)									
Detector distance (mm)			150						
Wavelength (Å)			1.54179						

	Diffraction data statistics of				
	lysozyme crystals in CDM				
2	crystalliza	ation plate			
	Crystal 26	Crystal 27			
	Crystar 20	Crystar 27			
Possibilition range (Å)	50-2.38	50-2.72			
Kesolution range (A)	(2.42-2.38)	(2.77-2.72)			
Mosaicity (°)	0.99	0.58			
< <i>I>/<σ(I)</i> >	16.68 (2.72)	6.29 (2.56)			
Wilson Plot B factor	27.10	25.40			
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2			
Call dimensions	<i>a</i> = 78.45	<i>a</i> = 76.83			
Cell unitensions	<i>b</i> = 78.45	<i>b</i> = 76.83			
<i>a</i> , <i>b</i> , <i>c</i> (Å)	2(21	27.11			
α, β, γ (°)	c = 30.21	c = 37.11			
	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$			
Exposure time (min)		5			
Oscillation angle (°)		1			
Detector distance	1	50			
(mm)	1.	50			
Wavelength (Å)	1.54	4179			

Supplementary	Table	S4. 2	X-ray	diffraction	data	statistics	of 23	lysozyme	crystals	in	SDVD
crystallization p	olate in	sam	e size								

	Diffraction data statistics of lysozyme crystals in SDVD crystallization plate				
2	Crystal 1	Crystal 2	Crystal 3	Crystal 4	Crystal 5
Deselution range (Å)	50-1.86	50-1.86	50-1.88	50-1.88	50-1.91
Resolution range (A)	(1.89-1.86)	(1.89-1.86)	(1.91-1.88)	(1.91-1.88)	(1.94-1.91)
Mosaicity (°)	0.61	0.91	0.75	0.82	0.71
< <i>I>/<</i> σ(<i>I</i>)>	18.51 (2.04)	15.43 (2.49)	11.83 (2.70)	13.74 (2.13)	14.69 (2.01)
Wilson Plot B factor	18.60	18.60	24.90	28.30	22.90
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2
	<i>a</i> = 78.49	<i>a</i> = 77.36	<i>a</i> = 75.79	<i>a</i> = 77.78	<i>a</i> = 76.30
Cell dimensions	<i>b</i> = 78.49	<i>b</i> = 77.36	<i>b</i> = 75.79	<i>b</i> = 77.78	<i>b</i> = 76.30
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 37.21	<i>c</i> = 37.52	<i>c</i> = 36.61	<i>c</i> = 37.54	<i>c</i> = 36.65
α, β, γ (°)	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$
Exposure time (min)			5		
Oscillation angle (°)			1		
Detector distance (mm)			150		
Wavelength (Å)			1.54179		

	Diffraction data statistics of lysozyme crystals in SDVD crystallization plate				
?					
	Crystal 6	Crystal 7	Crystal 8	Crystal 9	Crystal 10
Resolution range	50-1.93	50-2.00	50-2.02	50-2.03	50-2.03
(Å)	(1.96-1.93)	(2.03-2.00)	(2.05-2.02)	(2.06-2.03)	(2.06-2.03)
Mosaicity (°)	0.85	0.95	0.60	0.54	0.94
< <i>I>/<σ(I)></i>	10.35 (2.33)	21.78 (2.37)	13.51 (2.07)	13.10 (2.02)	15.47 (2.68)
Wilson Plot B factor	20.60	20.10	23.70	21.90	22.60
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2
Cell dimensions	<i>a</i> = 78.73	<i>a</i> = 78.87	<i>a</i> = 76.99	<i>a</i> = 77.97	<i>a</i> = 77.69
a h c(Å)	<i>b</i> = 78.73	<i>b</i> = 78.87	<i>b</i> = 76.99	<i>b</i> = 77.97	<i>b</i> = 77.69
a, b, c (A)	<i>c</i> = 37.99	<i>c</i> = 37.26	<i>c</i> = 37.11	<i>c</i> = 37.85	<i>c</i> = 37.85
α, ρ, γ ()	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$
Exposure time			5		
(min)					
Oscillation angle			1		
(°)					
Detector distance			150		
(mm)					
Wavelength (Å)			1.54179		

	Diffraction data statistics of lysozyme crystals in SDVD crystallization plate				
2	0 (111	0 + 112	0 + 112	0 + 114	0 + 115
	Crystal 11	Crystal 12	Crystal 13	Crystal 14	Crystal 15
Resolution range	50-2.08	50-2.10	50-2.12	50-2.15	50-2.24
(Å)	(2.12-2.08)	(2.14-2.10)	(2.16-2.12)	(2.19-2.15)	(2.28-2.24)
Mosaicity (°)	1.32	1.89	0.92	0.96	1.38
< <i>I>/<</i> σ(<i>I</i>)>	12.22 (3.18)	15.39 (6.07)	12.25 (2.26)	18.89 (5.41)	16.04 (2.48)
Wilson Plot B factor	23.50	12.80	22.00	21.80	26.00
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2
Cell dimensions	<i>a</i> = 76.44	<i>a</i> = 76.50	<i>a</i> = 75.87	<i>a</i> = 78.02	<i>a</i> = 79.00
$a \ b \ c(\text{\AA})$	<i>b</i> = 76.44	<i>b</i> = 76.50	<i>b</i> = 75.87	<i>b</i> = 78.02	<i>b</i> = 79.00
	<i>c</i> = 36.73	<i>c</i> = 37.62	<i>c</i> = 37.73	<i>c</i> = 37.48	<i>c</i> = 35.94
α, p, γ (*)	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$
Exposure time			5		
(min)					
Oscillation angle			1		
(°)					
Detector distance			150		
(mm)					
Wavelength (Å)			1.54179		

	Diffraction data statistics of lysozyme crystals in SDVD crystallization plate				
2	Crystal 16	Crystal 17	Crystal 18	Crystal 19	Crystal 20
Resolution range	50-2.26	50-2.28	50-2.45	50-2.51	50-2.61
(Å)	(2.30-2.26)	(2.32-2.28)	(2.49-2.45)	(2.55-2.51)	(2.66-2.61)
Mosaicity (°)	1.04	1.12	0.74	0.85	1.45
<1>/<\sigma(1)>	10.16 (3.49)	18.87 (2.69)	9.40 (2.05)	12.20 (2.78)	7.11 (3.35)
Wilson Plot B factor	24.40	24.40	27.40	28.50	32.90
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2
	<i>a</i> = 76.48	<i>a</i> = 75.70	<i>a</i> = 76.90	<i>a</i> = 76.01	<i>a</i> = 76.54
Cell dimensions	<i>b</i> = 76.48	<i>b</i> = 75.70	<i>b</i> = 76.90	<i>b</i> = 76.01	<i>b</i> = 76.54
<i>a</i> , <i>b</i> , <i>c</i> (A)	<i>c</i> = 37.01	<i>c</i> = 36.21	<i>c</i> = 37.50	<i>c</i> = 37.01	<i>c</i> = 36.97
α, β, γ (°)	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$
Exposure time			5		
(min)					
Oscillation angle (°)			I		
Detector distance (mm)			150		
Wavelength (Å)			1.54179		

	Diffraction data s	statistics of lysozym	e crystals in SDVD		
2	crystallization plate				
	Crystal 21	Crystal 22	Crystal 23		
Resolution range (Å)	50-2.80	50-3.25	50-3.45		
	(2.85-2.80)	(3.31-3.25)	(3.51-3.45)		
Mosaicity (°)	0.95	1.48	1.45		
< <i>I>/<σ(I)></i>	13.61 (2.62)	10.31 (73.05)	10.51 (2.34)		
Wilson Plot B factor	30.90	31.20	29.80		
Space group	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2	P4 ₃ 2 ₁ 2		
Cell dimensions	<i>a</i> = 75.73	<i>a</i> = 75.54	<i>a</i> = 76.08		
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>b</i> = 75.73	<i>b</i> = 75.54	<i>b</i> = 76.08		
α, β, γ (°)	<i>c</i> = 36.49	<i>c</i> = 36.54	<i>c</i> = 37.30		
	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$	$\alpha = \beta = \gamma = 90$		
Exposure time (min)		5			
Oscillation angle (°)		1			
Detector distance (mm)		150			
Wavelength (Å)		1.54179			

	Diffraction data statistics of ribonuclease A I crystals in CDM crystallization plate				
?					
-	Crystal 1	Crystal 2	Crystal 3	Crystal 4	Crystal 5
Resolution range	50-1.84	50-2.33	50-2.69	50-2.88	50-2.88
(Å)	(1.87-1.84)	(2.37-2.33)	(2.74-2.69)	(2.93-2.88)	(2.93-2.88)
Mosaicity (°)	0.95	1.29	1.85	1.28	1.04
< <i>I>/<</i> σ(<i>I</i>)>	13.31 (5.0)	13.37 (2.51)	9.29 (2.01)	9.80 (3.45)	7.36 (3.33)
Wilson Plot B factor	13.40	33.40	20.30	24.00	21.20
Space group	<i>P</i> 6 ₂ 22	P6 ₂ 22	P6 ₂ 22	<i>P</i> 6 ₂ 22	<i>P</i> 6 ₂ 22
	<i>a</i> = 63.63	<i>a</i> = 64.02	<i>a</i> = 66.16	<i>a</i> = 63.68	<i>a</i> = 63.59
Cell dimensions	<i>b</i> = 63.63	<i>b</i> = 64.02	<i>b</i> = 66.16	<i>b</i> = 63.68	<i>b</i> = 63.59
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 63.35	<i>c</i> = 64.21	<i>c</i> = 69.54	<i>c</i> = 64.36	<i>c</i> = 64.49
α, β, γ (°)	$\alpha = \beta = 90$	$\alpha = \beta = 90$	$\alpha = \beta = 90$	$\alpha = \beta = 90$	$\alpha = \beta = 90$
	<i>γ</i> = 120	$\gamma = 120$	$\gamma = 120$	$\gamma = 120$	$\gamma = 120$
Exposure time			10		
(min)					
Oscillation angle			1		
(°)					
Detector distance			150		

Supplementary Table S5. X-ray diffraction data statistics of 8 ribonuclease A I crystals in CDM crystallization plate in same size

(mm)

Wavelength (Å)

1.54179

Values in parentheses are for the highest resolution shell.

Continued Supplementary Table S5

	Diffraction data statistics of ribonuclease A I crystals in CDM crystallization plate				
2					
	Crystal 6	Crystal 7	Crystal 8		
Resolution range	50-3.16	50-3.18	50-3.52		
(Å)	(3.21-3.16)	(3.23-3.18)	(3.58-3.52)		
Mosaicity (°)	1.36	1.28	2.28		
< <i>I>/<</i> σ(<i>I</i>)>	5.88 (2.07)	7.26 (2.13)	6.26 (2.36)		
Wilson Plot B factor	28.10	30.80	33.00		
Space group	P6 ₂ 22	P6 ₂ 22	P6 ₂ 22		
	<i>a</i> = 63.51	<i>a</i> = 63.61	<i>a</i> = 64.91		
Cell dimensions	<i>b</i> = 63.51	<i>b</i> = 63.61	<i>b</i> = 64.91		
<i>a</i> , <i>b</i> , <i>c</i> (Å)	<i>c</i> = 63.41	<i>c</i> = 64.08	<i>c</i> = 65.23		
α, β, γ (°)	$\alpha = \beta = 90$	$\alpha = \beta = 90$	$\alpha = \beta = 90$		
	$\gamma = 120$	$\gamma = 120$	$\gamma = 120$		
Exposure time		10			
(min)					

Oscillation angle	1	
(°)		
Detector distance (mm)	150	
Wavelength (Å)	1.54179	

Values in parentheses are for the highest resolution shell.

Supplementary Table S6. The number of droplets containing amorphous precipitate in CDM and SDVD crystallization plates for the 11 different proteins.

Protein	Number of amorphous precipitate conditions		
	CDM	SDVD	
Proteinase K	15	11	
Lysozyme	19	16	
Ribonuclease A I	5	0	
Concanavalin A VI	17	12	
Catalase	13	11	
α-chymotrypsinogen A II	9	2	
α-chymotrypsin II	14	10	
Myoglobin	8	11	
Cellulase	6	4	
Hemoglobin	12	7	
Ribonuclease A III	10	3	